



10 December 2008

**APPLICATION FOR LICENCE FOR INTENTIONAL RELEASE OF GMOs INTO THE ENVIRONMENT: Application No. DIR 092**

**SUMMARY INFORMATION**

Project Title:	Limited and controlled release of wheat genetically modified to alter grain composition <sup>1</sup>
Applicant:	CSIRO
Common name of the parent organism:	Wheat
Scientific name of the parent organism:	<i>Triticum aestivum</i> L.
Modified trait(s):	Altered grain composition, antibiotic resistance
Identity of the gene(s) responsible for the modified trait(s):	<i>α</i> - and <i>γ</i> - <i>gliadin</i> from wheat <i>Starch Metabolic Enzyme I</i> <sup>2</sup> from wheat <i>Starch Metabolic Enzyme II</i> <sup>2</sup> from wheat <i>Starch Enzyme I</i> <sup>2</sup> from wheat <i>nptII</i> gene from the bacterial Tn5 transposon (antibiotic resistance)
Proposed Location(s):	One site in the ACT
Proposed Release Size:	One hectare
Proposed Release Dates:	July 2009 - June 2012

**Introduction**

The *Gene Technology Act 2000* (the Act) in conjunction with the *Gene Technology Regulations 2001*, an inter-governmental agreement and corresponding legislation that is being enacted in each State and Territory, comprise Australia's nationally consistent regulatory system for gene technology. Its objective is to protect the health and safety of people, and the environment, by identifying risks posed by or as a result of gene technology, and managing those risks by regulating certain dealings with genetically modified organisms (GMOs).

The Act establishes a statutory officer, the Gene Technology Regulator (the Regulator), to administer the legislation and make decisions under the legislation. The Regulator is supported by the Office of the Gene Technology Regulator (OGTR), an Australian Government regulatory agency located within the Health and Ageing portfolio.

The legislation sets out the requirements for considering applications for licences for dealings with GMOs and the matters that the Regulator must take into account before deciding whether, or not,

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<sup>1</sup> The title of the licence application submitted by CSIRO is 'Field trial of genetically modified wheat (*Triticum aestivum* L.) with altered grain composition'.

<sup>2</sup> CSIRO has sought approval to declare the precise identity of some genes as Confidential Commercial Information.

to issue a licence. The Regulator's *Risk Analysis Framework*<sup>3</sup> outlines the assessment process that will be followed.

## **The application and the proposed dealings**

The Acting Regulator has received an application from CSIRO for a licence for dealings involving the intentional release of genetically modified (GM) wheat (*Triticum aestivum* L.) into the Australian environment on a limited scale under controlled conditions.

Sixteen lines of GM wheat are proposed for release. The GM wheat contains gene silencing constructs designed to reduce expression of some genes involved in determining grain qualities important for dough making and nutritional purposes.

The purpose of the trial is to evaluate grain properties of the GM wheat lines grown under field conditions. This would involve generating sufficient grain to make flour for laboratory evaluation of how the flour performs in foods, and to feed to rats and pigs in laboratory experiments to determine whether altered grain properties change the nutritional value of the GM wheat.

The applicant proposes to limit the release to one site at a CSIRO research facility in the ACT, on a maximum area of 1 ha between July 2009 and June 2012. Access to the site would be limited to CSIRO staff only.

The applicant has also proposed a number of control measures to restrict the dissemination or persistence of the GM plants and their introduced genetic material that will be considered in the assessment of this application including:

- locating the trial site approximately 1 km away from natural waterways
- restricting animal access by surrounding the trial with a fence, mouse baiting the perimeter of the fence and covering the GMOs with bird-netting
- locating the trial site at least 200 m away from all other wheat plantings, with the exception of other GM trials, and at least 500 m away from plantings of wheat breeding lines
- minimising gene flow by surrounding the GM wheat with a 2 m wide buffer of non-GM wheat and preventing related species in the area surrounding the trial from flowering at the same time as the GMOs
- promoting the germination of any residual seed following harvest through three monthly cycles of irrigation and destroying any volunteer wheat with herbicide
- post harvest monitoring of the site for 24 months or until the site has been clear of volunteers for one growing season and destroying any volunteer wheat identified during this period
- destroying all plant materials from the trial site not required for testing or future trials
- transporting and storing the GMOs in accordance with OGTR guidelines
- not allowing the GM plant material or products to be used for human food or animal feed, with the exception of the above mentioned laboratory experiments, from which no material will enter the commercial human or animal food supply.

## **Confidential Commercial Information**

Some details, including names and sequences of genes are the subject of an application for declaration of Confidential Commercial Information (CCI) under section 185 of the Act, which is

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<sup>3</sup> More information on the assessment of licence applications is available from the Office of the Gene Technology Regulator (OGTR). Free call 1800 181 030 or at <<http://www.ogtr.gov.au>>.

currently under consideration. The confidential information will be made available to the prescribed experts and agencies that will be consulted on the Risk Assessment and Risk Management Plan (RARMP) for this application.

### **Parent organism**

The parent organism is bread wheat (*Triticum aestivum* L.) cultivars ‘Bobwhite 26’ and ‘NB1’. These cultivars are not grown commercially in Australia, but are commonly used in genetic modification work in Australia and overseas because they are relatively easy to transform.

Commercial wheat cultivation occurs in Australia’s wheat belt which extends from south eastern Queensland through New South Wales, Victoria, southern South Australia and southern Western Australia.

### **The genetic modifications and their effect**

The GM wheat lines contain gene constructs designed to reduce or silence the expression of specific wheat genes by a mechanism known as RNA interference (RNAi). These constructs are under the control of endosperm-specific promoters. These constructs target three gene families which, when silenced, change the composition of starch; and two gene families encoding gliadins, which are major seed storage proteins in wheat grains. Evaluation in contained facilities has shown that these lines have significantly altered grain composition. The GM wheat lines also contain a selectable marker gene (*nptII*) which confers resistance to aminoglycoside antibiotics related to kanamycin and neomycin.

Additionally, the GM wheat lines contain the *bla* gene from *Escherichia coli*, which confers resistance to the antibiotic ampicillin. This gene was used to select for bacteria containing plasmids<sup>4</sup> with the desired genes in the laboratory, prior to the production of the GM plants, and is not expressed in the GM wheat lines as it is linked to a bacterial promoter that does not function in plants.

Short regulatory sequences that control expression of the genes are also present in the GM wheat. These are derived from wheat, rice, Cauliflower mosaic virus (CaMV) and *Agrobacterium tumefaciens*. Although some of these sequences are derived from plant pathogens (CaMV and *Agrobacterium*), the regulatory sequences comprise only a small part of the pathogen’s total genomes, and are not in themselves capable of causing disease.

### **Method of genetic modification**

The 16 GM wheat lines proposed for release were derived from 15 independent transformation events, which were produced by two different methods. Fourteen of the lines for release were generated by transformation of the wheat cultivar Bobwhite 26 by the particle bombardment method. This involved ‘shooting’ wheat embryos with gold particles coated with two plasmids – one carrying the *nptII* plant selectable marker, the other carrying an RNAi construct designed to silence  $\alpha$ -gliadin,  $\gamma$ -gliadin, *Starch metabolic enzyme I* or *Starch metabolic enzyme II*. GM wheat plants were regenerated from the embryos using tissue culture techniques, including selection for the *nptII* marker gene. Molecular analysis was used to confirm presence of the RNAi construct.

Two of the GM wheat lines proposed for release are derived from conventional crosses between GM lines and carry two different RNAi constructs. These lines were generated by crossing *Starch metabolic enzyme II* RNAi lines described above with a wheat line genetically modified with an RNAi construct to silence *Starch enzyme I*. The *Starch enzyme I* RNAi construct, including the *nptII* gene and associated regulatory sequences, was originally introduced into the wheat cultivar NB1 on a plasmid vector carried by *Agrobacterium tumefaciens* (a common soil bacterium). The

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<sup>4</sup> Naturally occurring circles of DNA that are distinct and separate from the bacterial genome.

vector is 'disarmed' since it lacks the genes that encode the tumorigenic functions of *A. tumefaciens*.

### **Previous releases of the same or similar GMOs**

There has been no previous release of these GM wheat lines.

### **Suitability of Applicant**

Section 43(2)(f) of the Act requires the Regulator to be satisfied regarding the suitability of the applicant to hold a licence as a pre-requisite for considering DIR applications. The matters to be considered are outlined in Section 58 of the Act and include relevant convictions, revocation of a licence or permit relating to the health and safety of people, and capacity to meet the conditions of the licence.

The Acting Regulator has determined that CSIRO currently meets the suitability requirements and will verify this continues to be the case prior to making any decision regarding the issuing of a licence.

### **Consultation process for this DIR application**

The Acting Regulator has made an assessment of whether the application should be considered as a limited and controlled release, under section 50A of the Act. As its principal purpose is to enable the conduct of experiments, and the applicant has proposed limits on the size and duration of the release and controls to restrict the dissemination and persistence of both the GMO and its genetic material in the environment, **the Acting Regulator has decided that the application qualifies as a limited and controlled release.**

This means that the Acting Regulator is not required to consult on the assessment of this application until after a RARMP has been prepared in accordance with section 51 of the Act. In the interim, copies of the application are available on request from the OGTR. Please quote application number DIR 092.

The Acting Regulator will seek comment on the consultation RARMP from the public as well as a wide range of experts, agencies and authorities including the Gene Technology Technical Advisory Committee, State and Territory Governments, Australian Government agencies and the Minister for the Environment, Heritage and the Arts. The RARMP will then be finalised, taking into account matters raised relating to risks to human health and safety and the environment, and form the basis of her decision whether or not to issue a licence.

At this stage, **the RARMP is expected to be released for comment in mid March 2009.** The public will be invited to provide submissions on the RARMP via advertisements in the media and direct mail to anyone registered on the OGTR mailing list. The RARMP and other related documents will be available on the OGTR website, or in hard copy from the OGTR.

If you have any questions about the application or the assessment process, or wish to register on the mailing list, please contact the OGTR at:

**The Office of the Gene Technology Regulator, MDP54 GPO Box 9848 Canberra ACT 2601**

**Telephone: 1800 181 030 Facsimile: 02 6271 4202 E-mail: [ogtr@health.gov.au](mailto:ogtr@health.gov.au)**

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